REMARKS

Claim Status

Claims 1-45 are pending in the present Application. Claims 13-15 and 24-45 are withdrawn from consideration. Claims 1-12 and 16-23 are rejected. Claims 2-5, 9-12, 14, and 15 have been amended to clarify the claim language. Claims 16-21 have been amended to grammatically clarify the claim language. Claim 22 has been amended to clarify the claim language. No new matter has been added. Thus, entry and consideration of the amendments is respectfully requested.

OBJECTIONS

IDS

The previously-filed Information Disclosure Statement, filed February 22, 2007 is allegedly not in compliance with 37 CFR § 1.97, 1.98 and MPEP § 609. A supplemental IDS is being submitted under separate cover, to provide the additionally required information for some of the previously-listed documents, as required by the Examiner. Therefore, all documents submitted for consideration have been fully cited, and the objection has been overcome. The Applicants therefore request that the objection be withdrawn.

Claim Objections

Claims 18 and 21 are objected to as having grammatical errors. Claims 18 and 21 have been amended to recite "a flow cytometer detection system", thus obviating the objection. Claims 16, 17, 19, and 20 have been similarly amended for consistency. Therefore, the objection has been overcome and the Applicants respectfully request withdrawal of the objection.

REJECTIONS

Rejection Under 35 USC § 112, 2nd Paragraph

Claims 1-12 and 16-23 are rejected under 35 USC § 112, 2nd Paragraph as being indefinite.

Claim 1

The Examiner alleges that Claim 1 is an incomplete method claim because the claim recites measurements "in a biological sample", but that the method taught in the working example draws venous blood from each subject before and after oral feeding with a probiotic preparation, and then incubates PBMCs isolated from the blood of the subjects *in vivo*. Thus, the Examiner asserts that the cytokines are not measured "in a biological sample". Therefore, the Examiner asserts that the claimed method is not the method disclosed in the specification, and it is unclear what the claim is directed to. Dependent Claims 2-12 and 16-21 are similarly rejected as dependent on Claim 1.

The Applicants respectfully traverse the rejection. The Applicants assert that the method as recited in Claim 1 is consistent with the method described in the specification. The *in vivo* aspect of the method is that the patients are actually fed a potential treatment (i.e. a probiotic). The biological samples (the venous blood) are drawn before *and after* the three week treatment with the probiotic and are then compared. The PBMCs harvested from the blood can be further stimulated in the method, but need not be. See page 14, lines 17-20. Even if the PBMCs are further stimulated, they are still the PBMCs in the biological sample and their secretions are what is being measured before and after administration of a potential treatment which is administered *in vivo*. Therefore, the cytokines are measured in the biological sample from a mammalian subject. Note the contrast with the *in vitro* method in which the cells used in the *in vitro* method are taken from subjects not treated with a potential treatment. The treatment of the cells in the *in vitro* method is only after harvesting and culturing, whereas with the *in vivo* method, a biological sample from a treated subject is used.

Thus, the Applicants assert that Claim 1 is complete, is clear as to what the Claim is directed to, and is consistent with the method disclosed in the specification. Therefore,

the rejection has been overcome and the Applicants respectfully request withdrawal of the rejection with respect to Claim 1 and also Claims 2-12 and 16-23 which depend therefrom.

Claims 2-5, 9-12 16, 17, 18 and 20

Claims 2-5 are alleged to be vague and indefinite for reciting specific cytokines and "mixtures thereof", as the Examiner does not understand how many of the cytokines the Applicants intend to measure. The Examiner further asserts that the 'inflammatory cytokine' can not be a mixture of two or more cytokines.

The Applicants respectfully traverse the rejection. Claims 2-5, as amended, are in Markush format. Claiming in Markush format is a well understood and accepted claim format. If desired, one could measure levels and ratios of more than one pair of cytokines, and it is not required, in Markush claim format to specify every possible combination. Reciting "and combinations thereof" in the Claims, as amended, is not vague or indefinite. One of skill in the art would understand that levels of one, two or more cytokines could be measured, levels of all of the recited anti-inflammatory and pro-inflammatory cytokines could be measured, or levels of various combinations of cytokines could be measured, and ratios of the levels compared. See The Cytokine Handbook, page 1386, lines 6-9, wherein it is described that more than one cytokine can be simultaneously measured in the same cell supernatant. Claims 2-5 have been amended to clarify that various combinations of cytokines can be measured.

Beginning at page 11 of the Specification, the Applicants clearly describe that levels of *at least one* anti-inflammatory cytokine can be measured and the level compared to the level of *at least one* pro-inflammatory cytokine. The preferred cytokines from which to chose are described, as recited in amended Claims 2-5. It is also described that levels of any anti-inflammatory cytokine, as described in the Specification, can be compared to levels of any pro-inflammatory cytokine as described in the Specification. See page 12, lines 31-33 wherein particularly useful ratios of particular cytokines are described. Ratios of particular cytokines are believed to be particularly indicative of efficacy of treatment for

inflammatory bowel diseases, however, levels of any of the recited cytokines can be measured and compared.

Thus, Claims 2-5, as amended, are clear, and the rejection has been overcome. The Applicants therefore respectfully request withdrawal of the rejection.

Claims 9-11

Claims 9-11 are alleged to be vague and indefinite because the Examiner is not clear as to whether the Applicant intends to use all of the recited elements (biological samples) together or separately. The Examiner further alleges that a biological sample can not be a mixture of one or more biological samples.

The Applicants respectfully traverse the rejection. Claims 9-11, as amended, are Markush groups, written in an acceptable alternative format. Claiming in Markush format is a well understood and accepted claim format and it is not required, in Markush claim format, to specify every possible combination. Thus, it is clear that a biological sample can be obtained from any of the recited types of samples, that more than one of the sample types may be taken and tested, and that different combinations of samples can be obtained and tested. Claims 9-11 have been amended to clarify that various combinations of samples can be taken and tested.

Thus, Claims 9-11, as amended, are clear, and the rejection has been overcome. The Applicants therefore respectfully request withdrawal of the rejection.

Claim 12

Claim 12 is alleged to be vague and indefinite because the Examiner is not clear as to whether the Applicant intends all of the recited elements (compounds which stimulate PBMCs) are to be used together or separately, or whether the Applicants intend to use two or more elements at a time or sequentially.

The Applicants respectfully traverse the rejection. Claim 12, as amended, is a Markush group written in an alternative format. Markush style claiming is a well understood and accepted claim format. Thus, one of skill in the art would understand that the stimulation could occur with one or more of the members of the recited group, individually or in combination.

Therefore, the rejection has been overcome and the Applicants respectfully request that the rejection be withdrawn.

Claims 16, 17, 19, and 20

Claims 16, 17, 19, and 20 are alleged to be vague and indefinite because the Examiner asserts that it is unclear how one can have mixtures of different assay methods, and asserts that the word "mixture" is commonly understood to refer to a combination of different substances.

The Applicants respectfully traverse the rejections. Claims 16, 17, 19 and 20 have been amended. The Claims, as amended, recite that the means for measuring could be any of the members of the group, used individually or in combination – i.e. one could measure levels of cytokine using one or more than one method.

Therefore, Claims 16, 17, 19, and 20, as amended, are clear and the rejection has been overcome. Because the rejection has been overcome with respect to Claims 16, 17, 19, and 20, the rejection has also been overcome with respect to Claims 18 and 21 which depend therefrom. The Applicants therefore respectfully request withdrawal of the rejection.

Claim 22

Claim 22 is alleged to be vague and indefinite because the Examiner asserts that a kit, by definition, must contain two or more elements and the interrelationships between the elements must be explicitly stated. Claim 23 is also rejected as being dependent on Claim 22.

The Applicants respectfully traverse the rejection. Claim 22 does specify two elements: A kit comprising *a first measuring element or system* for measuring the level of at least one anti-inflammatory cytokine in a biological sample from a mammalian subject before treatment and at at least one time point after or during treatment, *a second measuring element or system* for measuring the level of at least one pro-inflammatory cytokine in a biological sample from said mammalian subject before treatment, and at at least one time point after or during treatment,...

Furthermore, Claim 22 does specify the interrelationship between the two measuring elements or systems: ... wherein the change in ratio of the level of anti-inflammatory to pro-inflammatory cytokine after administration of the treatment can be determined and usage instructions provided instructing the user to determine the ratio of the level of an anti-inflammatory cytokine to the level of a pro-inflammatory cytokine both before and after treatment, wherein an increase in said ratio of the level of said anti-inflammatory cytokine to the level of said pro-inflammatory cytokine is indicative of an inhibitor of inflammatory diseases of the bowel.

The interrelationship is that the first measuring element or system measures the level of at least one anti-inflammatory cytokine, the second measuring element or system measures the level of a pro-inflammatory cytokine, and the two levels are then compared in order to study the ratio between the two. Thus, the two measuring elements or systems are used and are related in that their results are then used to calculate the ratios.

Therefore, Claim 22 does posses the required two or more elements – the first and second measuring elements or systems, and the interrelationship between the two is explicitly stated in that the levels of cytokine determined by each measuring element or system are compared to calculate ratios. Thus, the rejection has been overcome and the Applicants respectfully request withdrawal of the rejection with respect to Claims 22 and 23.

Rejection Under 35 USC § 112, 1st Paragraph

Claims 1-12 and 16-21 are rejected under 35 USC § 112, 1st Paragraph for allegedly failing to comply with the enablement requirement.

Claims 1-12 and 16-21 are rejected because the Examiner asserts that only methods which measure cytokine levels in tissues directly from the bowel region, or methods which measure cytokine production by peripheral blood mononuclear cells with *in vitro* stimulation, gut lymphoid tissues witn *in vitro* stimulation, or gut lymphoid tissues without *in vitro* stimulation are enabled.

Additionally, the Examiner asserts that the application is not enabled for *all* treatments, particularly treatment that involves administering anti-inflammatory cytokines or compositions which interact directly with pro-inflammatory cytokines.

The Examiner further asserts that only methods which determine changes in the ratio of the levels of IL-10/IL-12, IL-10/TNF-α, and IL-10/IFN-γ after treatment as indicative of efficacy of treatment are enabled.

The Examiner reasons that due to the allegedly large quantity of experimentation necessary to determine that changes in any cytokine level in any biological sample would be indicative of an efficacy of treatment, alleged lack of direction/guidance presented in the specification regarding same, the absence of working examples regarding same, the complex nature of the invention, the state of the art which establishes that some treatments of IBD comprise direct treatment with cytokines, and that assays of cytokine levels in some biological samples are often not meaningful, and the breadth of the claims which encompass any treatment modality and assasy utilizing any biological sample, undue experimentation would be required of the skilled artisan to make and/or use the claimed invention.

The Applicants respectfully traverse the rejections. The test of enablement is whether one reasonably skilled in the art could make or use the invention from the disclosures in

the patent coupled with information known in the art without undue experimentation. See MPEP § 2164.01 citing *United States v. Telectronics, Inc.*, 857 F.2d 778, 785, 8 USPQ 2d 1217, 1223 Fed. Cir. 1988. The fact that experimentation may be complex does not necessarily make it undue, if the art typically engages in such experimentation. See MPEP § 2164.01 citing *In re Certain Limited-Charge Cell Culture Microcarriers* 221 USPQ 1165, 1174, Int'l Trade Comm'n 1983, *aff'd. sub nom., Massachusetts Institute of Technology v. A.V. Fortia,* 774 F.2d 1104, 227 USPQ 428 Fed. Cir. 1985.

As noted by the Examiner, the factors to be considered to determine whether experimentation is "undue" include the breadth of the claims, the nature of the invention, the state of the prior art, the level of one of ordinary skill, the level of predictability in the art, the amount of direction provided by the inventor, the existence of working examples, the quantity of experimentation needed to make or use the invention based on the content of the disclosure. See MPEP § 2164.01(a).

The Applicants assert that undue experimentation would not be required by one of skill in the art in order to practice the invention, and that the Claims are enabled. The Applicants provide sufficient background for one of skill in the art to understand the problems associated with inflammatory bowel diseases, and the methods and technologies described in the present Specification. In addition, the level of skill of one in the art would be relatively high as the invention involves scientific and medical principles. Furthermore, complex experimentation is common to the biological and medicinal arts. The Applicants assert that based on the description and definitions provided in the Specification, including example anti-inflammatory cytokines, pro-inflammatory cytokines, types of samples that can be tested, types of stimulating agents that can be used *in vitro* and *in vivo,* and the presence of the working example, one of skill in the art would be able to understand and make or use the invention.

In particular, the Applicants assert that methods beyond those which measure only cytokine levels in tissues directly from the bowel region, methods which measure cytokine production by peripheral blood mononuclear cells with *in vitro* stimulation, gut

lymphoid tissues witn *in vitro* stimulation, or gut lymphoid tissues without *in vitro* stimulation are enabled. The Examiner cites The Cytokine Handbook, pages 1384 -1386 for the proposition that measurements of plasma levels of cytokines are often not meaningful. Thus, the Examiner asserts that the Claims are not enabled for tissues other than those directly from the bowel region. However, sentence the Examiner cites continues with the exception: "... when studying pharmacokinetics of exogenously administered cytokines *or in conditions associated with chronic cytokine release*". Thus, if IBS results in chronic, altered cytokine ratios, IBS may be a condition associated with chronic cytokine release and may be studied with methods measuring cytokine levels in tissues other than those directly from the bowel region. Therefore, the Applicants assert that the Claims are enabled for all of the claimed tissues and methods.

Additionally, the Examiner asserts that the application is not enabled for *all* treatments, particularly treatment that involves administering anti-inflammatory cytokines or compositions which interact directly with pro-inflammatory cytokines. The Applicants respectfully traverse the rejection. Even though such treatments may be known, the methods of the Application can still be used to study the subject's response to the treatment, and to determine the efficacy of the treatment. Therefore, the Applicants assert that the Claims and application are enabled for all treatments.

The Examiner further asserts that only methods which determine changes in the ratio of the levels of IL-10/IL-12, IL-10/TNF-α, and IL-10/IFN-γ after treatment as indicative of efficacy of treatment are enabled. The Applicants respectfully traverse the rejection. As noted in The Cytokine Handbook cited by the Examiner, cytokine profiles are known, and levels of multiple cytokines can be measured from the same cell supernatant. In addition, at page 1384, line 45 to page 1385, line 3 of the Cytokine Handbook, it is described that multiplex assay kits can measure multiple cytokines in a single sample. In addition, as the Examiner notes, it is known that there are numerous cytokines that have pro and anti-inflammatory properties. Thus, one of skill in the art would understand from the working example that other pro and anti-inflammatory cytokines could be used and

studied. Thus, it would be well within the ability of one of skill in the art to determine and study various cytokine ratios without undue experimentation.

The Applicants therefore assert tha the Claims and application are enabled broadly for pro and anti-inflammatory cytokines and ratios thereof. Therefore, the rejection has been overcome and the Applicants respectfully request withdrawal of the rejection.

Rejections Under 35 USC § 102

Claim 22 is rejected under 35 USC § 102(b) as allegedly being anticipated by Vignali, 2000 Journal of Immunological Methods 243:243-255 (hereafter "Vignali").

The Examiner asserts that Claim 22, given its broadest reasonable interpretation, requires a product for measuring multiple cytokines in a biological sample from a mammalian subject. The Examiner asserts that there are no structural limitations recited as to the contents of the kit. Therefore, the Examiner asserts that ratio calculation steps do not convey patentability to the kit; that because there are no structural limitations in the Claim, a kit of the Claim could include various components of a FlowMetrix™ system as disclosed in Vignali; and that where the only difference between a prior art product and a claimed product is printed matter that is not functionally related to the product, the content of the printed matter will not distinguish the claimed product from the prior art.

The Applicants respectfully traverse the rejection. Under 35 USC §102, anticipation requires that all the Claim elements appear in a single prior art document. "A Claim is anticipated only if each and every element set forth in the Claim is found, either expressly or inherently described, in a single prior art reference." MPEP § 2131 citing *Verdegal Bros. v. Union Oil Co. of California*, 814 F.2d 628, 631, 2 USPQ2D 1051, 1053 (Fed. Cir. 1987). "The identical invention must be shown in as complete detail as contained in the ... Claim." MEPE § 2131 citing *Richardson v. Suzuki Motor Co.*, 868 F.2d 1226, 1236, 9 USPQ2D 1913, 1920 (Fed. Cir. 1989).

Vignali discloses that the simultaneous detection of various analytes can be achieved using the FlowMetrix™ assay. Vignali discloses that multiple cytokine levels can be

measured simultaneously. Vignali does not disclose a kit. Vignali simply uses an assay method to measure various analytes and discusses the advantages and disadvantages of various assay methods and devices. No kit is described in Vignali, regardless of whether specific structures are recited for the kit of Claim 22. In addition, Claim 22 recites providing instructions to a user instructing the user in determining desired ratios. Vignali does not disclose providing instructions to users of a kit. Furthermore, the instructions provided in the kit, instructing the user in determining desired useful ratios are functionally related to the kit product and its components, and would be different than any instructions included with the systems discussed in Vignali.

Therefore, all elements of the Claim are not found in Vignali, and Vignali can not, as a matter of law, anticipate the Claim. The Applicants, therefore, respectfully request that the rejection be withdrawn.

35 USC § 103

Claim 23 is rejected under 35 USC § 103(a) as being allegedly unpatentable over Vignali.

The Examiner acknowledges that Vignali does not disclose a kit comprising means for collecting biological samples. However, the Examiner asserts that it would have been obvious for one of ordinary skill in the art to include a device for obtaining a sample, that one would be motivated because it would increase the efficiency utilization of an assay to determine cytokine levels.

The Applicants respectfully traverse the rejection. The Examiner has not established a *prima facie* case of obviousness, even in light of *KSR International Co. v. Teleflex Inc.* 82 USPQ2d 1385 (U.S. 2007) ("KSR"). See also MPEP § 2143.01. KSR did not eliminate the need for at least some suggestion, motivation, or expectation of success for making a given modification. A *prima facie* case of obviousness has not been established because the cited document does not teach or suggest all of the claim limitations of Claim 23, or provide any reasonable motivation or expectation of success for trying the presently claimed invention. See MPEP § 2143.03.

The Examiner asserts that Vignali discloses a kit for measuring cytokines in a biological sample. Nowhere does Vignali disclose a kit. Vignali only discloses various assay methods and devices, and compares them. Thus, there is no teaching or suggestion to provide a kit. Thus, because there is no teaching or suggestion to provide a kit at all, there is no motivation to add to such a kit a means for collecting biological samples. Even if one of skill in the art understood that a biological sample would need to be obtained, such information would not have led one of skill in the art to the present invention. Vignali simply performs various assays on various equipment to test the limits, practicality, cost, advantages and disadvantages of various assays done on various equipment. There is nothing in Vignali that teaches or suggests a kit. Thus, there is no expectation of success found in Vignali for adding to a kit a means for collecting biological samples.

Therefore, Vignali does not provide the requisite teaching, suggestion, motivation, or expectation of success to have led one of ordinary skill in the art to the present invention, and does not disclose every element of the present invention. Thus, the rejection has been overcome and the Applicants respectfully request that the rejection be withdrawn.

Claims 1-5, 9, 16, 17, 19, and 20 are rejected under 35 USC § 103(a) as being allegedly unpatentable over Towaga et al. 2002 Am J. Physiol, Gastrointestinal Liver Physiol 283:G187-G195 ("Towaga").

The Examiner asserts, although Towaga does not teach or suggest measuring the levels of anti and pro-inflammatory cytokines before administering a treatment, measuring cytokine levels in tissue biopsies, or determining the ratio of levels of anti to pro-inflammatory cytokines, that it would have been obvious for one of ordinary skill to measure levels before and after treatment, that one would have been motivated to make the jump from animal model experiments to actual clinical experimentation, and that one would have been motivated to investigate and analyze ratios of levels of cytokines.

The Applicants respectfully traverse the rejection. The Examiner has made too great a leap. Towaga details a particular experiment in rats using induced colitis. Towaga does not provide any suggestion to study 'before and after' results, or to set up such experiments. Towaga does not teach or suggest studying ratios of cytokines to establish and analyze shifts in patterns of cytokine levels. Towaga simply induces colitis in rats and compares cytokine levels to those of normal, control rats in conjunction with studying physical aspects of the induced disease such as thickness of the colon, weight of the colon, and presence and size of lesions, in order to determine whether lactoferrin is effective against the induced colits. There is no teaching or suggestion to do completely different clinical experiments, or to use or analyze 'before and after' data in a clinical setting. Simply because one could do such experiments and such analysis is not sufficient, even in light of *KSR*. There still must be some reason or motivation to choose a particular experiment or analysis and Towaga does not provide such motivation, particularly with the wide range of possible experiments and analyses that are available to be done when studying various disease conditions.

Therefore, the disclosure of Towaga, in combination with the knowledge of one skilled in the art does not teach the claimed invention, and one of skill in the art would not have been led to the claimed invention based on the disclosure of Towaga. Therefore, the rejection has been overcome and the Applicants respectfully request that the rejection be withdrawn.

Claims 18 and 21 are rejected under 35 USC § 103(a) as being allegedly unpatentable over Togawa as applied to Claims 1, 17 and 20 in view of Vignali. The Examiner acknowledges that Togawa does not teach a method of measuring levels of at least one anti-inflammatory cytokine and at least one pro-inflammatory cytokine in a biological sample by multiplexed ELISAs using coded microspheres coupled with a flow cytometer detection system. However, The Examiner asserts that Vignali teaches a FlowMetrix™ system that uses microspheres as the solid support for a conventional immunosorbent assay. Therefore, the Examiner asserts that it would have been obvious to modify the

teachings of Togawa and substitute the multiplex assay taught by Vignali for the ELISA assay taught by Togawa.

The Applicants respectfully traverse the rejection. Even if Towaga were combined with Vignali and a FlowMetrix™ system were used, the claimed invention would not result. Perhaps one could analyze the cytokine levels of Towaga with such a system. However, one would not have arrived at the claimed method of determining the efficacy of a treatment of inflammatory diseases of the bowel in mammals *in vivo*. Neither Towaga nor Vignali teaches or suggests a clinical method, using samples from a biological subject, in which cytokine levels are determined and ratios analyzed. Neither Towaga nor Vignali contemplates such a method.

In addition, Claims 18 and 21 depend ultimately from Claim 1 which is novel and non-obvious over the cited documents. Therefore Claims 18 and 21 are also novel and non-obvious over the cited documents.

Therefore, the rejection has been overcome and the Applicants respectfully request that the rejection be withdrawn.

Claims 6-8 are rejected under 35 USC § 103(a) as being allegedly unpatentable over Togawa as applied to Claim 1 in view of Blumberg et al. 1999 Current Opinion in Immunology 11:648-656 ("Blumberg"). The Examiner acknowledges that Togawa does not teach the particular claimed ratios of cytokines. However, the Examiner asserts that Togawa teaches measurement of various cytokines, and that Blumberg teaches the importance of the balance of particular cytokines in IBD pathogenesis. Therefore, the Examiner asserts that it would have been obvious to modify the teachings of Togawa and substitute measurement of pro-inflammatory cytokines as taught by Blumberg. The Examiner also asserts that calculation of ratios would have been obvious as a way of monitoring changes in the balance of levels of pro and anti-inflammatory cytokines.

The Applicants respectfully traverse the rejection. As noted above, Towaga does not teach or suggest establishing or analyzing any ratios of cytokines. Blumberg also does not teach or suggest establishing or analyzing ratios. Blumberg simply notes that there is likely an on-going balance between pro and anti-inflammatory cytokines, and their release and activity in body systems in relation to inflammation. Blumberg is simply a review of known animal models of mucosal inflammation and their potential relation to human inflammatory bowel disease. Blumberg summarizes which animal models might be better for studying various types of inflammatory bowel disease such as Ulcerative Colitis and Crohn's Disease. Blumberg does not teach methods for evaluating efficacy of treatments. Therefore, even if one were to have combined the teachings of Towaga and Blumberg, one would not have arrived at the Applicants' invention.

In addition, Claims 6-8 depend from Claim 1 which is novel and non-obvious over the cited documents. Therefore Claims 6-8 are also novel and non-obvious over the cited documents.

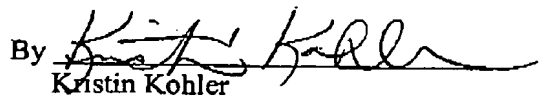
Therefore, the rejection has been overcome and the Applicants respectfully request withdrawal of the rejection.

Conclusion

The Applicants therefore respectfully request that Examiner reconsider the Application. Early allowance of all pending Claims is respectfully requested. If the Examiner believes that personal contact would be beneficial for disposition of the present application, the Examiner is respectfully requested to contact the undersigned.

Respectfully submitted,

THE PROCTER & GAMBLE COMPANY

By 
Kristin Kohler
Registration No. 41,907
(513) 662-3371

Date: November 28, 2007
Customer No. 27752